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Influence of Diet on Chick Growth-Promoting and Antiperotic Properties of Betaine, Methionine and Choline.*

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Results showing that betaine is effective in preventing perosis and promoting growth in chicks fed a simplified diet (Diet 543) have been reported by McGinnis, Norris and Heuser.[1] The improved growth was found to be highly significant. Jukes and Welch[2] reported that betaine had a growth promoting effect on chicks fed a purified diet, but was without effect in preventing perosis. Almquist and Grau[3] using a purified diet containing isolated soybean protein instead of casein and gelatin reported that betaine and methionine were as effective as choline in promoting growth. In later work Almquist and Grau[4] with a somewhat different basal diet obtained a growth-promoting effect from betaine only when combined with cystine or homocystine. This effect, however, was not equal to that obtained with choline.

In order to explain the discrepancy between the results of McGinnis and associates,[1] Jukes and Welch,[2] and Almquist and Grau,[3, 4] 3 experiments are here reported showing the influence of betaine, methionine and choline on perosis and growth in chicks when added to basal diets differing in composition. Day-old Rhode Island Red cockerel chicks were used in all of the experiments, 12 per lot in Experiment 1 and 15 per lot in Experiments 2 and 3. The experiments were terminated at the end of 4 weeks.

The composition of the experimental diets is given in Table I. Diet 543 was found to contain 0.078% choline, diet 661 to contain 0.027%, and diet 671 to contain 0.040%. The approximate methionine content of diet 661 was 0.85% and the cystine content 0.09%, and the approximate methionine content of diet 671 was 0.38% and the cystine content 0.07%. From a consideration of the available data, it appears probable that the approximate methionine content of diet 543 was 0.55% and the cystine content 0.20%. The data used in obtaining these values were for the most part those reported by Schmidt,[5] Grau and Almquist,[6] and Brown.[7]

The results of the experiments are given in Table II. In Experiment I choline and betaine were used as supplements to diets 543 and 661. The original findings of McGinnis and associates[1] that betaine when added to simplified diet 543 prevents perosis and promotes growth were confirmed. On the other hand, betaine had no effect on perosis or growth when added to purified diet 661. Its ineffectiveness on perosis is in agreement with that obtained by Jukes and Welch,[2] with a purified diet. In contrast to the results with betaine, choline prevented perosis and pro-

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1 McGinnis, J., Norris, L. C., and Heuser, G. F., PROC. SOC. EXP. BIOL. AND MED., 1942, **51**, 293.

2 Jukes, T. H., and Welch, A. D., *J. Biol. Chem.*, 1942, **146**, 19.

3 Almquist, H. J., and Grau, C. R., *J. Biol. Chem.*, 1943, **149**, 575.

4 Almquist, H. J., and Grau, C. R., *J. Nutrition*, 1944, **27**, 263.

5 Schmidt, C. L. A., *The Chemistry of the Amino Acids and Proteins*, 1944, 2nd ed., 217.

6 Grau, C. R., and Almquist, H. J., *J. Nutrition*, 1943, **26**, 631.

7 Brown, W. L., *J. Biol. Chem.*, 1942, **142**, 299.

TABLE I.
Composition of Basal Diets.

Ingredient	Diet 543	Diet 661	Diet 671
	g	g	g
Degerminated yellow corn meal	66.75	—	—
Cerelose	—	52.0	52.0
Peanut meal	15.00	—	—
Purified casein	10.00	25.0	—
Isolated soybean protein	—	—	25.0
Gelatin	—	5.0	5.0
Dried brewers' yeast	—	5.0	5.0
Mineral mixture	5.00*	5.0†	5.0†
Corn oil	—	4.0	4.0
Soybean oil	3.00	—	—
Cellophane	—	3.0	3.0
Fortified cod liver oil‡	0.25	1.0	1.0
	mg	mg	mg
Inositol	—	100.0	100.0
p-Aminobenzoic acid	—	10.0	10.0
Niacin	—	3.0	3.0
Pantothenic acid	0.7	1.5	1.5
Alpha-tocopherol	—	1.0	1.0
2-Methyl-1,4-naphthoquinone	—	1.0	1.0
Riboflavin	0.5	1.0	1.0
Pyridoxine	0.5	0.5	0.5
Thiamin	0.3	0.5	0.5
Biotin	—	0.01	0.01

* *Poultry Science*, 1940, **19**, 315.

† Bone meal, 2270 g; K_2HPO_4, 840 g; pulverized limestone, 700 g; NaCl, 600 g; $MgSO_4 \cdot 7H_2O$, 500 g; $Fe_2(SO_4)_3 \cdot XH_2O$, 55 g; $MnSO_4 \cdot 4H_2O$, 29 g; KI, 3.5 g; $CuSO_4 \cdot 5H_2O$, 1.5 g; $ZnCl_2$, 1.0 g; $CoCl_2 \cdot 6H_2O$, 0.2 g.

‡ 2000 U.S.P. units vitamin A and 400 A.O.A.C. units vitamin D per g.

moted growth when added to either diet 543 or diet 661.

In Experiment 2 methionine was used as a supplement to diets 543 and 661 as well as betaine and choline. Again betaine was as effective as choline in preventing perosis and promoting growth when added to simplified diet 543. In agreement with the results of Experiment 1, betaine failed to prevent perosis or promote growth when added to purified diet 661. Methionine was as effective in promoting growth as betaine or choline when added to diet 543 and markedly reduced the incidence and severity of perosis. Like betaine, methionine was ineffective in preventing perosis or promoting growth when added to diet 661.

From a comparison of the results on these diets, it is evident that betaine and methionine when added to diet 543 enable the chick to synthesize choline and that choline is required as such for the prevention of perosis and for growth. The effectiveness of betaine and methionine in preventing perosis and promoting growth when added to diet 543 may be attributed to the presence of an unidentified factor necessary for the synthesis of choline by the chick from these substances. The possibility, however, that a factor which inhibits choline synthesis is present in diet 661, cannot be excluded at the present time.

In experiment 3 additional data were obtained which support the conclusion that choline is required as such for the prevention of perosis and for growth in chicks. Diet 671 fed in this experiment was the same as purified diet 661 except that a fraction of the proteins of soybeans, isolated by special procedures, replaced the purified casein. With possibly one exception the growth response

TABLE II.
Effect of Choline,* Betaine,* and Methionine on Perosis and Growth in Chicks.

Diet	Supplement, 0.2% each	Perosis		Avg wt at 4 wks with st
		Incidence %	Severity index	
	Exp. 1.			
543	None	72.5	37.4	195 ± 31.6
	Choline	0.0	0.0	277 ± 46.1
	Betaine	0.0	0.0	266 ± 34.2
661	None	100.0	57.0	151 ± 19.6
	Choline	0.0	0.0	252 ± 72.3
	Betaine	100.0	68.0	150 ± 28.6
	Exp. 2.			
543	None	84.7	38.9	179 ± 59.6
	Choline	0.0	0.0	231 ± 35.8
	Betaine	6.6	0.7	232 ± 45.7
	Methionine	26.6	8.1	227 ± 59.4
	Choline + methionine	0.0	0.0	234 ± 35.7
	Betaine + "	7.7	1.3	205 ± 35.3
661	None	80.0	37.8	187 ± 32.7
	Choline	0.0	0.0	218 ± 55.8
	Betaine	100.0	52.0	174 ± 33.2
	Methionine	93.0	57.2	165 ± 27.0
	Choline + methionine	0.0	0.0	224 ± 68.5
	Betaine + "	93.0	58.8	151 ± 28.3
	Exp. 3.			
671	None	71.4	29.4	82 ± 14.2
	Choline	0.0	0.0	169 ± 30.3
	Betaine	100.0	56.5	124 ± 21.3
	Methionine	100.0	48.8	111 ± 20.2
	Choline + betaine	0.0	0.0	179 ± 33.3
	Choline + methionine	0.0	0.0	210 ± 39.7
	Betaine + "	100.0	56.7	147 ± 27.5
	Choline + betaine + methionine	0.0	0.0	240 ± 40.0

* Added as the chloride.
† Standard deviation.

obtained with choline was approximately the same regardless of whether it was added to diet 671 alone or in combination with betaine, or methionine, or betaine and methionine. Betaine and methionine were effective in promoting growth but not in preventing perosis. The growth results with betaine are in general agreement with those obtained by Almquist and Grau[4] under somewhat different conditions, and by Jukes and Welch[2] using a purified diet of lower methionine content than diet 661.

The discrepancy in the results obtained with diets 661 and 671 is probably due to the fact that the latter diet contained less methionine and hence fewer methyl groups than the former. The methyl-group deficiency is also revealed by comparing the difference in growth response obtained by supplementing those diets with betaine and methionine. Maximum growth and also prevention of perosis on diet 671 were not obtained however, until choline was included in it along with betaine and methionine. The results of this work are in general agreement with those of Almquist and Grau[4] who reported that the growth-promoting effects of betaine or methionine and arsenocholine were additive under their experimental conditions.

Summary. Betaine and methionine have been shown to be effective in preventing perosis and promoting growth in chicks when added to a simplified diet. They were ineffective when added to a purified diet of higher

methionine content. In contrast, choline prevented perosis and promoted growth on both diets.

The results indicate that choline is required as such for the prevention of perosis and for growth and that betaine and methionine when added to the simplified diet enabled the chick to synthesize choline. This may be caused by an unidentified factor present in the simplified diet which is necessary for choline synthesis. Another possible explanation is that a factor which inhibits choline synthesis is present in the purified diet.

When the casein in the purified diet was replaced with an isolated fraction of the proteins of soybeans with resultant reduction in methionine content, betaine and methionine failed to prevent perosis but promoted some growth. The growth response obtained with betaine and methionine in this instance is believed to be due to a deficiency of methyl groups only. Choline was again effective for both purposes.

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Initial Emptying Time of Stomach in Primigravidae as Related to Evacuation of Biliary Tract.*

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Previous studies[1] have shown that in the second and third trimesters of pregnancy the gall bladder of 13 gravidæ discharged an average of only 49% of contents (within 40 minutes after a standard meal of egg-yolk) as compared with an average discharge of 73% obtained from 12 nulligravidæ; that the best 8 of the 13 discharged an average of only 57% and the remaining 5 an average of only 38%; but when the latter were tested 6 to 8 weeks postpartum, the amount of bile discharged in the 40-minute period (71%) approximated the nulligravid figure.

The present study has attempted to ascertain whether this retardation in rate of emptying of the gall bladder in pregnancy could have been due to a delay in the initial rate of emptying of the stomach. To this end 17 nulligravidæ (nursing students) were examined fluoroscopically after receiving a standard meal of egg-yolk to which 3 heaping teaspoons of barium sulphate had been added. They were then used as controls for 18 primigravidæ of the second and third trimesters, similarly treated. Fluoroscopic observations were supplemented by films. In addition to recording the time of the first spurt into the bulb, and the length of time required for the head of the meal to reach the pars inferior duodeni and the jejunum, an attempt was made to estimate the amount discharged from the stomach during the first 15 minutes. After the above tests were concluded, it was ascertained by experiment that 10 cc of egg-yolk (2/5 of a swallow) was enough to induce evacuation of 72% of the contents of the gall bladder in 35 minutes, when injected directly into the duodenum.

Table I summarizes the most striking results. In each of the 2 groups of subjects there were 13 individuals in whom the pylorus opened within the first few minutes.

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1 Gerdes, Maude M., and Boyden, Edward A., *Surg., Gyn., and Obst.*, 1938, **66**, 145.